Test Definition: VHPV

Human Papillomavirus (HPV) Vaginal Detection with Genotyping for High-Risk Types by PCR

Overview

Useful For

Detection of high-risk (HR) genotypes associated with the development of cervical cancer

Aids in triaging women with abnormal Pap smear results

Individual genotyping of human papillomavirus (HPV)-16 and/or HPV-18 if present

Results of HPV-16 and HPV-18 genotyping can aid in triaging women with positive HR-HPV but negative Pap smear results

This testing is intended for use in clinical monitoring and management of patients. It is **not intended for** use in medical-legal applications.

This test is **not intended for** use in determining the need for treatment (ie, excisional or ablative treatment of the cervix) in the absence of high-grade cervical dysplasia. Patients who are HPV16/18 positive should be monitored carefully for the development of high-grade cervical dysplasia according to current practice guidelines.

Method Name

Real-Time Polymerase Chain Reaction (PCR)

NY State Available

Yes

Specimen

Specimen Type

Vaginal

Necessary Information

Specimen source, collection date and patient identifiers are required.

Specimen Required

Patient Preparation: For 24 hours prior to specimen collection, patients **should not use** carbomer-containing feminine hygiene products (eg, KY Jelly [Physician Formula], Vagisil creme regular strength). For additional information about carbomer-containing feminine hygiene products, including additional examples, see Cautions.
Specimen Type: Vaginal
Container/Tube: ThinPrep/PreservCyt solution vial
Specimen Volume: 3 mL of solution in ThinPrep/PreservCyt vial

Collection Instructions:

1. Obtain an adequate sampling from vagina using a sterile polyester tipped swab.
2. Rinse the swab as quickly as possible into the PreservCyt Solution vial.
3. DISCARD THE SWAB.
4. Bag ThinPrep specimens individually as they have a tendency to leak during transport.
2. Place labels on the vial and on the bag.

Forms

If not ordering electronically, complete, print, and send a Microbiology Test Request (T244) with the specimen.

Specimen Minimum Volume

1 mL

Reject Due To

Specimen containing CytoRich Red preservative fluid	Reject

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Vaginal	Ambient (preferred)	42 days	
	Refrigerated	42 days	

Clinical & Interpretive

Clinical Information

Persistent infection with human papillomavirus (HPV) is the principal cause of cervical cancer. The presence of HPV has been implicated in more than 99% of cervical cancers worldwide, including both cervical squamous cell carcinoma and cervical adenocarcinoma. Before the development of invasive cancer, HPV infects the squamous mucosa cells and/or the glandular cells of the endocervix, leading to clonal expansion and morphologic changes. While the HPV-infected cells are restricted to their normal anatomic location, these changes are classified as cervical intraepithelial neoplasia (CIN). The severity of the morphologic changes and the degree to which those changes resemble the morphology of an invasive carcinoma are used to "grade" CIN. In general, high-grade CIN more closely resembles invasive carcinoma morphologically. HPV can also infect other mucosal cells in the anogenital region, such as the vaginal mucosa, leading to the development of HPV-associated intraepithelial neoplasia as well as invasive carcinoma not involving the cervix itself, although this is less common.

Human papillomavirus is a small, nonenveloped, double-stranded DNA virus, with a genome of approximately 8000 nucleotides. There are more than 118 different types of HPV and approximately 40 different HPVs can infect the human

anogenital mucosa. Only a very small percentage of patients who are exposed to HPV will develop CIN. Of those patients who develop CIN, only a small percentage will progress to invasive cervical cancer. Sexually transmission of HPV is extremely common, with estimates of up to 75% of all women being exposed to HPV at some point. However, almost all infected women will mount an effective immune response and clear the infection within 2 years without long-term health consequences. Both high-risk HPV genotypes (especially HPV-16 and 18), as well as persistent HPV infection (eg, an infection that is not cleared by the patient's immune system over time), are associated with an increased chance of progressing to high-grade CIN and invasive cancer.

Data suggest that certain HPV genotypes (eg, HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68) are high-risk (HR) for the development of cervical cancer and its precursor lesions. Furthermore, HPV types 16 and 18 have been regarded as the genotypes most closely associated with progression to cervical cancer. HPV-16 is the most carcinogenic and is associated with approximately 60% of all cervical cancers, while HPV-18 accounts for approximately 10% to 15% of cervical cancers.

In developed countries with cervical cancer screening programs, the Pap smear has been used since the mid-1950s as the primary tool to morphologically detect CIN, the precursor to cervical cancer. Pap smear screening has decreased death rates due to cervical cancer dramatically, since in many cases CIN can be treated and eliminated (eg, by local excision) before it progresses to invasive carcinoma. Although Pap smears and other liquid-based cytology methods have many advantages, they also have limitations: they require subjective interpretation by a highly trained cytopathologist and misinterpretation can occur, morphologic changes that resemble HIV-associated CIN can be caused by other conditions (eg, inflammation), and Pap smear does not sample every cell within the cervix/anogenital region potentially leading to falsely negative results. Perhaps most importantly, Pap smear does not differentiate between HPV genotypes that are high or low risk for progression to cervical cancer and it does not detect very early infections, which may lack a morphological phenotype.

Nucleic acid (DNA) testing by polymerase chain reaction has become a standard, noninvasive method for determining the presence of a cervical HPV infection. Proper implementation of nucleic acid testing for HPV may:
1) increase the sensitivity of cervical cancer screening programs by detecting high-risk lesions earlier in women 30 years and older with normal cytology.
2) reduce the need for unnecessary colposcopy and treatment in patients 21 and older with cytology results showing atypical squamous cells of undetermined significance.

Data suggest that individual genotyping for HPV types 16 and 18 can assist in determining appropriate follow-up testing and triaging women at risk for progression to cervical cancer. Studies have shown that the absolute risk of CIN-2 or worse in HPV-16 and/or HPV-18 positive women is 11.4% (95% confidence interval [CI] 8.4%-14.8%) compared with 6.1% (95% CI, 4.9%-7.2%) of women positive for other HR-HPV genotypes, and 0.8% (95% CI, 0.3%-1.5%) in HR-HPV-negative women. Based in part on these data, the American Society for Colposcopy and Cervical Pathology now recommends that HPV 16/18 genotyping be performed on women who are positive for HR-HPV but negative by routine cytology. Women who are found to be positive for HPV-16 and/or -18 may be referred to colposcopy, while women who are negative for genotypes 16 and 18 may have repeat cytology and HR-HPV testing in 12 months.

Reference Values

Negative for human papillomavirus (HPV) genotypes 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68

Interpretation

A positive result indicates the presence of human papillomavirus (HPV) DNA from one or more of the following genotypes: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68.

For patients with atypical squamous cells of undetermined significance Pap smear result and who are positive for high-risk (HR) HPV, consider referral for colposcopy, if clinically indicated.

A negative result indicates the absence of HPV DNA of the targeted genotypes.

For women aged 30 years and older with a negative Pap smear test result but who are positive for HPV-16 and/or HPV-18, consider referral for colposcopy, if clinically indicated.

For women aged 30 years and older with a negative Pap smear test result, positive-HR-HPV test result, but who are negative for HPV-16 and HPV-18, consider repeat testing by both cytology and a HR-HPV test in 12 months.

Cautions

Patients should refrain from using carbomer-containing feminine hygiene products for 24 hours prior to collection. Use of these products has been associated with invalid results of the HPV detection/genotyping assay. Carbomer-containing products include, but are not limited to:

-KY Jelly (Physician Formula)
-Surgilube Surgical Lubricating Jelly
-HR lubricating Jelly
-DynaLybe Lubricating Jelly
-McKesson Lubricating Jelly
-Cardinal Health Lubricating Jelly
-Medline Lubricating Jelly
-Conceptrol Contraceptive Gel
-Labicam anti-fungal
-Lavena Moisturizer
-Monistat 1
-Terrasil Ointment Plus Cleansing Bar
-VCF Contraceptive Foam
-Walgreens Clotrimazole 3
-Walgreens Clotrimazole Vaginal Cream
-RepHresh Clean Balance
-RepHresh Vaginal Gel prefilled
-Vagisil anti-itch cream
-Vagisil creme regular strength
-Vagisil ProHydrate
-Vagisil Sensitive Cream
-IsoLove Balancing Gel
-Replens Long-Lasting Vaginal Moisturizer
-Metronidazole Vaginal Gel

The cobas human papillomavirus (HPV) test is US Food and Drug Administration (FDA)-approved for cervical and endocervical samples collected in PreservCyt (ThinPrep) media. Other sample types (eg, vaginal) are not considered FDA-approved sources; however, verification studies have been completed by Mayo Clinic Laboratories and Mayo Clinic in compliance with CLIA regulations.

The cobas HPV test detects DNA from high-risk genotypes 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68. This test does not detect DNA of HPV low-risk types (eg, 6, 11, 42, 43, 44) since these are not associated with cervical cancer and its precursor lesions.

Prevalence of HPV infection in a population may affect performance. Positive-predictive values decrease when testing populations with low prevalence or individuals with no risk of infection.

Infection with HPV is not an indicator of cytologic high-grade squamous intraepithelial lesion (HSIL) or underlying high-grade cervical intraepithelial neoplasia (CIN), nor does it imply that CIN2-3 or cancer will develop. Most women infected with 1 or more high-risk (HR) HPV types do not develop CIN2-3 or cancer.

A negative-HR-HPV result does not exclude the possibility of future cytologic HSIL or underlying CIN2-3 or cancer.

Cervical specimens often show visibly detectable levels of whole blood as a pink or light brown coloration. These specimens are processed normally on the cobas systems. If concentrations of whole blood exceed 10% (dark-red or brown coloration) in PreservCyt solution, there is a likelihood of obtaining a false-negative result.

The cobas HPV Test performance has not been validated with PreservCyt specimens that have been treated with up to 5% glacial acetic acid for removal of red blood cells. Addition of glacial acetic acid over 5% in PreservCyt specimens prior to HPV testing would invalidate the cobas HPV Test results.

Human beta-globin amplification and detection is included in cobas HPV to differentiate HPV negative specimens from those that do not exhibit HPV signal due to insufficient cell mass in the specimen. All HPV negative specimens must have a valid beta-globin signal within a pre-defined range to be identified as valid negatives.

The cobas HPV Test performance has not been validated with PreservCyt specimens that have been filled past the maximum fill line of the primary vial. ThinPrep vials that have had any additional PreservCyt fluid volume added or any dissimilar fluid volume added to the initial specimen should not be submitted for testing.

The presence of polymerase chain reaction inhibitors may cause false-negative or invalid results.

Human papillomavirus-negative cancers of the cervix do occur in rare circumstances. Also, no cancer screening test is 100% sensitive. Use of this device for primary cervical cancer screening should be undertaken after carefully considering the performance characteristics put forth in the cobas HPV Test label, as well as recommendations of professional guidelines.

The use of this test has not been evaluated for the management of women with prior ablative or excisional therapy, hysterectomy, who are pregnant or who have other risk factors (eg, HIV-positive, immunocompromised, history of sexually transmitted infections).

The effects of other potential variables (eg, vaginal discharge, use of tampons, and douching), and specimen collection variables have not been evaluated.

Clinical Reference

1. Saslow D, Solomon D, Lawson HW, et al. American Cancer Society, American Society for Colposcopy and Cervical Pathology, and American Society for Clinical Pathology screening guidelines for the prevention and early detection of cervical cancer. J Low Genit Tract Dis. 2012;16(3):175-204. doi:10.1097/LGT.0b013e31824ca9d5
2. Walboomers JM, Jacobs MV, Manos MM, et al. Human papillomavirus is a necessary cause of invasive cervical cancer worldwide. J Pathol. 1999;189(1):12-19. doi:10.1002/(SICI)1096-9896(199909)189:1
3. de Sanjose S, Quint WG, Alemany L, et al. Human papillomavirus genotype attribution in invasive cervical cancer: a retrospective cross-sectional worldwide study. Lancet Oncol. 2010;11(11):1048-1056. doi:10.1016/S1470-2045(10)70230-8
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5. US Preventive Services Task Force, Curry SJ, Krist AH, et al. Screening for cervical cancer: US Preventive Services Task Force Recommendation Statement. JAMA. 2018;320(7):674-686. doi:10.1001/jama.2018.10897
6. Poljak M, Balencak AO, Cuschieri K, Bohinc KB, Arbyn M. 2023 global inventory of commercial molecular tests for human papillomaviruses (HPV). J Clin Viro. 2024:172;105671
7. Perkins RB, Guido RS, Castle PE, et al. 2019 ASCCP Risk-Based Management Consensus Guidelines for abnormal cervical cancer screening tests and cancer precursors. J Low Genit Tract Dis. 2020;24(2):102-131. doi:10.1097/LGT.0000000000000525

Performance

Method Description

The cobas HPV (human papillomavirus) test is a qualitative real-time polymerase chain reaction (PCR) test that detects 14 high-risk HPV genotypes. The test uses primers to define a sequence of approximately 200 nucleotides within the polymorphic L1 region of the HPV genome. A pool of HPV primers present in the Master Mix is designed to amplify HPV DNA from 14 high-risk types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68). An additional primer pair targets the human beta-globin gene (330 base pair amplicon) as an internal control to monitor the entire sample preparation and PCR amplification process. Fluorescent oligonucleotide probes bind to polymorphic regions within the sequence defined by these primers. The test utilizes a low titer positive and a negative control.(Package insert: cobas HPV: Qualitative nucleic acid test for the cobas 5800/6800/8800 Systems. Roche Diagnostics, Inc; Rev. 2.0, 09/2024)

PDF Report

No

Day(s) Performed

Monday through Saturday

Report Available

3 to 6 days

Specimen Retention Time

2 weeks

Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Superior Drive

Fees & Codes

Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification

This test has been modified from the manufacturer's instructions. Its performance characteristics were determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information

87626
G0476 (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
VHPV	HPV Vaginal Detect / Genotyping PCR	77378-8

Result ID	Test Result Name	Result LOINC® Value
619403	HPV High Risk type 16, PCR	61372-9
619404	HPV High Risk type 18, PCR	61373-7
619405	HPV other High Risk types, PCR	77375-4